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RESEARCH ARTICLE OPEN ACCESS

Bacterial Colonization of Intensive Care Unit Environment and Healthcare Workers in A Tertiary Care Hospital in Kolar Region, India

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Abstract

Direct shedding of microbes by patients and health care workers results in contamination of Intensive care unit environment. Intensive care unit acquired infections due to microbial contamination is a major concern because the patient's immunity is already compromised. To determine the rate of bacterial contamination on environmental surfaces of Intensive care unit and health care workers and to determine the antibiogram of the isolates. Air samples and swabs from healthcare workers, their accessories, surrounding environmental surfaces were collected randomly over a period of 2 months in Adult Intensive care units. Bacterial isolates were identified by standard microbiological techniques. Antibiotic sensitivity testing was performed by Kirby Bauer disc diffusion method and data analyzed by Statistical Product and Service Solutions 22 version software. A total of 208 samples were randomly collected over 2 months, of which 56 samples yielded positive bacterial growth. Of 56 growth, 12 isolates were detected from air sampling method and 44 isolates from swabs. Among 44 isolates identified from swabs, 10 were isolated from healthcare workers, 4 from health care worker's accessories and 30 from environmental surfaces. Six different bacterial isolates were identified, Coagulase Negative Staphylococcus (24) and Micrococcus (15) were the major isolates followed by Non fermenters (6), *Staphylococcus aureus*(4), *Bacillus* species(4) and diphtheroids (3) The antimicrobial sensitivity pattern of these bacterial isolates were sensitive to commonly used antibacterial agents. Study results showed Intensive care unit staff and environmental surfaces as probable sources of bacterial contamination. Study highlights the importance of cleaning and disinfection process and educate the health care workers about the possible sources of infections within Intensive care unit.

Keywords: Bacterial contamination, Intensive care unit, antibiotic sensitivity

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INTRODUCTION

Intensive care unit (ICU) is an essential element of effective health care services that caters the care of resuscitation, management and monitoring of critically ill patients[1]. The microbial contamination of intensive care units is of a major concern as it can contribute to ICU acquired infections. Breach in the body barriers due to invasive devices, surgery, co-morbid conditions like diabetes, hypertension etc and prolonged antimicrobial exposure compromise patient's immunity admitted in ICU[2].

In health care facilities, ICU acquired infections accounts for more than 20% of nosocomial infections[3]. The clinical activities in ICU involves use of higher antibiotics,minor procedures, invasive monitoring, and managing critically ill patients, all these factors favour the emergence of multidrug resistant bacterial strains resulting in high morbidity and mortalityrate[4].

The contaminated environmental surfaces play a significant role in the transmission of healthcare associated pathogens[5]. In ICU patients are surrounded by equipment for monitoring the vitals and organ support like ventilator, infusion pumps and resuscitation trolleys[6]. The contamination of ICU environment and medical devices may occur as the consequence of cross transmission or direct shedding of microbes by patient or by healthcare workers (HCWs)[7]. In addition, the ICU staff can serve as vehicles for the spread of resident pathogens among patients in ICU's[8].

Periodic surveillance of ICU is essential to ascertain the level of bacterial contamination on environmental surfaces and HCW's. The study was conducted to determine the rate of bacterial contamination on environmental surfaces in Intensive care unit and health care workers and to determine the antibiogram of the isolates.

MATERIALS AND METHODS

Study Setting, design and period

Across-sectional descriptive study was conducted from July-August 2019 at Adult Intensive Care Unit of R L Jalappa Hospital and Research Centre,a tertiary care teaching hospital in rural Kolar region, Karnataka, India Air Samples and swabs from HCW's, their accessories and environmental surfaces was collected randomly once in 2 weeks over 2 months period in Adult intensive care unit of 30 bedded capacity. A total of 4 rounds of surveillance was conducted over the study period two months duration accounting for a total of 208 samples (52 samples in each round).

In each round of surveillance, 20 samples from environmental surfaces, 12 samples from anterior nares and hands of HCWs (two Intensivists, two Nurses and two housekeeping staff) 15 from HCWs accessories (mobile, pen, stethoscopes and rings) and 5 from air samples i.e a total of 52 samples were collected.

Swabs from the both right and left anterior nares and hands of ICU HCWs (2 Intensivists, 2 Nurses and 2 housekeeping staff) and their accessories (mobile, pen, stethoscopes and rings) were collected. Swab samples were chosen randomly and collected from ICU environmental surfaces and devices that were in close contact with the patients. Twenty swabs were collected from floor, walls,door handles, taps, bed linens, iv sets, beds, cot, nursing station, trolleys, ambu bags,oxygen masks, ventilators, suction machine, sphygmomanometer, table, chairs, patient files, telephone handsets and stethoscopes.

Collection and Processing

Air sample

Air sampling was done by settle plate method. Open Blood Agar and MacConkey agar plates, prelabeled with site area were placed at 5 areas (4 corners and 1 at the centre) of the ICU about1 meter above the ground, 1 meter from the wall and exposed for 1 hour following the schedule 1/1/1[9]. Plates were transported to the Microbiology laboratory within 10 minutes at room temperature. These plates were incubated at 37°C for 24 to 48 hours in microbiology laboratory, was observed for any bacterial growth.

Swab sample

Prelabeled sterile swab were moistened in sterilesaline and was rolled over the inanimate surfaces, equipment's, HCWs hands, anterior nares and the nit was transported to the microbiology laboratory within 10 minutes at room temperature. The swabs were immediately streakedon to Blood agar and MacConkey agar media and incubated at 37°C for 24 -48 hours in microbiology laboratory, was observed for any bacterial growth.

Any bacterial growth was further identified using standard bacteriological methods

and appropriate biochemical tests carried out based on the standard operating procedure (like gram stain, catalase, coagulase, oxidase, indole, citrate, urease, mannitol motility and triple sugar iron tests)[10,11].

Antibiotic susceptibility testing

Susceptibility testing was done on Mueller Hintonagar for all isolates by Kirby Bauer disc diffusion method according to the latest CLSI guidelines. Cefoxitin disc was used for the screening of Methicillin Resistant *Staphylococcus aureus* (MRSA)[12].

Quality contro

Escherichiae coli ATCC 25922, *Klebsiella pneumoniae* ATCC 700603, *Pseudomonas aeruginosa* ATCC 27853, *Staphylococcus aureus* ATCC 25923 and *Enterococcus faecalis* ATCC29212 were used for Internal qualitycheck.

Statistical Methods

Data was entered into Microsoft excel data sheet and was analysed using Statistical Product and Service Solutions (SPSS)22Version software.

Ethical Approval

The study protocol was approved by the Institutional Ethical committee of Sri Devaraj Urs Medical College. (Reference no SDUMC/KLR/IEC/287/2019-20). The HCWs were included in the study after obtaining informed participant consent

RESULTS

A total of 208 samples were collected from 4 rounds of surveillance, of which 56 (26.92 %) samples yielded positive bacterial growth. Of these 56-positive growth, 12 (21.36%) were

Table 1. Distribution of bacteria on the various sites in ICU

Site	*S.aureus*	CONS	Micrococcus	Bacillus	Diphtheroids	NFGNB	TOTAL
Settle plate		5	6	1			12 (21.36)
HCW Nasal swab	3	6			1		10 (17.8)
HCW mobile			2				2 (3.56)
HCW rings		2					2 (3.56)
Door handle		1					1 (1.78)
ICU tap				1		2	3 (5.34)
Bed linen			1		1		2 (3.56)
IV set stand		1					1 (1.78)
Cot		1				1	2 (3.56)
Nursing station		1					1 (1.78)
Trolley medication			1				1 (1.78)
Ambu bags		1	1				2 (3.56)
Oxygen mask		1					1 (1.78)
Suction apparatus		1					1 (1.78)
Telephone set			1				1 (1.78)
Patient Files					1		1 (1.78)
Stethoscope			1				1 (1.78)
Multipara monitor				1			1 (1.78)
Handwash sink	1					1	2 (3.56)
Ultrasound			1				1 (1.78)
ABG Analyzer		1					1 (1.78)
ECG Leads						2	2 (3.56)
Computer mouse		1					1 (1.78)
Computer keyboard			1				1 (1.78)
Dressing trolley		1					1 (1.78)
Refrigerator				1			1 (1.78)
Camera monitor		1					1 (1.78)
Total	4(7.14)	24(42.72)	15 (26.7)	4 (7.14)	3 (5.34)	6(10.71)	56 (100)

HCWs - Health care workers; CONS - Coagulase negative Staphylococcus; *S.aureus - Staphylococcus aureus*
NFGNB - Non fermenting Gram negative bacilli; Numerical value in the bracket denotes percentage

isolated from Air sampling method and 44 (78.57 %) by swab samples.

Among 44 (78.57 %) positives growth from swabs, 10 (17.8%), 4 (7.14%) and30 (53.4%) were isolated from HCW's, their accessories and environmental surfaces respectively as shown in Table 1.

Six different bacterial isolates were identified. Coagulase Negative *Staphylococcus* (CONS) 24 (42.72%) and *Micrococcus* 15 (26.7%) accounted for majority of the isolates followed by Non fermenters 6 (10.71%)*Staphylococcus aureus* 4 (7.14%), Bacillus 4 (7.14%) and diphtheroids 3 (5.34%).

Bacterial contamination of ICU from environmental surfaces, inanimate objects, health care workers and air quality are represented in Table 1.

CONS 24 (42.72%) and Micrococcus 15(26.7%) accounted for high contamination rate. More than 1 isolate was observed from ICU taps, handwash sink, ECG leads, ambu bags, cot, bed linen as evidenced in the Table 1.

No microbial contamination was detected on the following objects –IV infusion sets, bed railings, ventilators, sphygmomanometer, dialysis machine, defibrillator, walls and partition curtains.

The antibiogram of Gram positive and Gram negative bacterial isolates are shown in the Table 2 and 3 respectively. The isolates were sensitive to most of the commonly used Antibacterial agents. No Multidrug resistant bacteria isolated in our study.

DISCUSSION

ICU acquired infections accounts for major health problem globally leading to higher morbidity and mortality. The potential sources of ICU infections are patient's flora (40-60%) followed by health care workers and their accessories (20-40%) and contaminated environmental surfaces and equipment (20%)[13].

The prevalence of ICU acquired infections in developed countries is around 5–10%, while their prevalence exceeds 2–20 times higher in developing countries[14].

The results of our study showed higher contamination of the environmental surfaces and medical devices by Gram positive 24 (80%) when compared to Gram negative 6 (20%) types. Gram positive bacteria were predominantly comprised of Coagulase negative *Staphylococcus* followed by *Micrococcus*, *Bacillus*, Diphtheroids and *Staphylococcus aureus*. Non fermenters were isolated among Gram negative bacteria. Our findings were concurrent with a study conducted by Tajeddin et al. which showed contamination is more with Gram positive than Gram negative (60.7% v/s 39.3 %)[15]. This may be due to the better survival of Gram positive bacteria in contrast to the Gram negative bacteria in dryair[16,17].

Table 2. Antibiotic sensitivity pattern of Gram positive bacteria isolated from ICU

Antibiotics	*S.aureus* N = 4	CONS N =24	Micrococcus N =15	Bacillus N= 4	Diphtheroids N = 3
Penicillin	0 (0)	18 (82.8)	12(79.9)	4 (100)	3 (100)
Erythromycin	2 (50)	22 (99.52)	12(79.9)	3(75)	3 (100)
Ciprofloxacin	2 (50)	20 (83.2)	13 (86.5)	4 (100)	2 (66.6)
Cefazolin	3(75)	22 (99.52)	15 (100)	4 (100)	3 (100)
Gentamicin	3(75)	22 (99.52)	14 (93.2)	3(75)	3 (100)
Cotrimoxazole	3(75)	20 (83.2)	13 (86.5)	3(75)	2 (66.6)
Tetracycline	3(75)	23 (95.68)	15 (100)	4 (100)	3 (100)
Chloramphenicol	4 (100)	24 (100)	15 (100)	4 (100)	3 (100)
Amoxycillin clavulanic acid	4 (100)	24 (100)	15 (100)	4 (100)	3 (100)
Linezolid	4 (100)	24 (100)	15 (100)	4 (100)	3 (100)
Vancomycin	4 (100)	24 (100)	15 (100)	4 (100	3 (100)

Numerical in the Bracket denotes percentage %

Table 3. Antibiotic sensitivity pattern of Gram negative bacteria isolated from ICU

Antibiotics	NFGNB (N = 6)
Cotrimoxazole	1 (16.66)
Ciprofloxacin	1 (16.66)
Ceftazidime	1 (16.66)
Levofloxacin	1 (16.66)
Doxycycline	2 (33.33)
Amikacin	3 (50)
Cefepime	3 (50)
Tobramycin	4 (66.66)
Piperacillin	4 (66.66)
Piperacillin Tazobactum	6 (100)
Imipenem	6 (100)
Meropenem	6 (100)

In contrast to our findings, a study done by Jadhav et al. reported that Gram negative bacteria contributed a major proportion on the ICU inanimate objects than Gram positive (68.8 %v/s 31.1%)[18]. This may be due to the intrinsic resistance exhibited by Gram negative bacteria to disinfectants as their cell wall is impermeable to active biocide agents and they also possess degradative enzymes[19].

Indoor air contamination of ICU accounts for 10-33% of Nosocomial infections[20]. Our study showed indoor air quality surveillance assessed by Air sampling method yielded Coagulase Negative *Staphylococcus* 6 (50%) followed by *Micrococcus* 5 (41.6%), and *Bacillus* 1(8.3%). These isolates could be commensal flora of human skin, mucus membranes that are continuously shred[21,22].

A study conducted by Kiranmai et al. showed ICU air sample was highly contaminated with bacteria like *Bacillus* Spp as well as potential pathogens like *Klebsiella, Pseudomonas*[23]. No potential pathogens were isolated from our study. Our study showed lesser air contamination of 12 (21.3%) with *Micrococcus* and CONS as predominant isolates which could be commensal flora. This could be attributed to lesser movement of staff members and restriction of visitors and better infection control practices. The high level of air contamination in other studies may be attributed to the movement of large equipment, increased movement of staff numbers, bed changes, patient personal hygiene, increase visiting hours and inadequate cleaning[24].

Several studies document that hands of ICU staff accounts for 20-40% infections due to the cross transmission between colonised / infected patients[25,26]. A study conducted by Tajeddin et al. on the hands of ICU staff yielded *Acinetobacter baumannii* (1.4%), *Staphylococcus aureus* (5.9%), *epidermidis* (20.9%) and *Enterococcus* spp. (1%)[15]. In contrast, our study did not show any evidence of bacterial colonization from the HCWs hand swabs collected before performing procedures. This can be attributed to compliance of our HCWs to hand hygiene practices.

There are several studies confirming the nasal colonization of MRSA as a major risk factor for infections in ICU caused by the colonizing strain[27-29]. The study conducted by Joanchim et al. showed the prevalence of MRSA carriers among HCWs was 59/379 (15.6%)[30]. A study conducted by Warnke et al. revealed that bacterial detection depends on the uptake and release capacities of the swabs and the swabbing techniques[31]. In our study, screening of the Nasal swabs of the health care workers (Intensivists / Nursing staff/ housekeeping staff) didn't show any evidence of MRSA colonization, but showed the presence of *Staphylococcus aureus* (8.92%), Coagulase Negative Staphylococcus (10.71%) and diphtheroids (1.78%)

The accessories (Stethoscopes, mobiles, pens) used by the HCWs for the improvement of the patients may pose a major threat to patients admitted in ICU[32,33]. A study conducted by Lavanya et al. showed that 56% of mobile phones, 52% of the stethoscopes, 40% of finger rings, and 28% of the pens used by ICU staff showed growth of coagulase negative *Staphylococcus* spp. (70.46%), *Staphylococcus aureus* (13.69%) and *Acinetobacter* spp (11.64%)[34]. In our study it was observed that the accessories of HCWs yielded CONS(3.56%) and *Micrococcus* (3.56%). This could be attributed to strict infection control practices by our ICU staff.

The ICU sinks are used for cleaning hands and medical equipment before disinfection and to flush patient's secretions / fluids. These activities can induce biofilm production and emergence of multidrug resistance and acts as a potential source[35].

A study conducted by Geyter et al. revealed that the sink in the ICU was a potential source of infection resulting in many outbreaks of Carbapenemase producing *Enterobacteriaceae*[36]. Another study conducted by Kramer et al. found that 100% of the sinks in an ICU were contaminated with Gram negative bacilli due to several non-hand hygiene activities[37]. In our study, ICU sink and the tap showed lesser contamination and yielded Non fermenters and *Staphylococcus aureus* which accounted for only8.92%.

The antibiotic sensitivity pattern of our isolates showed low rates of drug resistance in contrast with other studies where they have reported high resistant pattern[38,39]. None of our isolates were resistant to reserve antibiotics like vancomycin, linezolid, piperacillin tazobactum and carbapenems. Our study did not yield any MDR isolates, may be due to the strict adherence of our Intensivists to antibiotic policies/antimicrobial stewardship program. The high resistance pattern reported by other studies may be attributed to the selective pressure due to extensive use of broad-spectrum antibiotics[40].

CONCLUSION

Our study results showed ICU staff as well as environmental surfaces as probable sources of bacterial contamination. Bacterial contamination can contribute to ICU acquired infections. HCWs should be aware of the risk of cross-transmission of microbes between them to inanimate surfaces and vice versa. The hospital infection control and prevention team should conduct periodic surveillance, effective cleaning of environmental surfaces, sterilizing the instruments before and after use, and strictly adhere to basic standard precautions at all the times during health care activities.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

AUTHORS' CONTRIBUTION

KR helped in Collection and processing of samples, collection of data. AN helped in designed of the study, processing of the samples, analysis and interpretation of sample, analysing data and writing the paper. SKN and DK helped in Revision of intellectual contents, interpretations of samples, manuscript editing. MMR helped in statistical analysis and editing of manuscript.

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DATA AVAILABILITY

All datasets generated or analyzed during this study are included in the manuscript.

ETHICS STATEMENT

The institutional ethics committee of Sri Devaraj URS medical college, tamaka, kolar has examined and unanimously approved this study.

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